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BRIEF COMMUNICATION

OBSERVATIONS ON THE BREEDING BIOLOGY OF A MICROHYLID FROG (GENUS *OREOPHRYNE*) FROM NEW GUINEA

The conquest of terrestrial environments is a major theme in vertebrate evolution. Classically the Amphibia have been viewed as an intermediate stage between the aquatic and terrestrial groups[1,2]. However, the standard view of the amphibian life-cycle involving eggs hatching into aquatic tadpoles which subsequently metamorphose into terrestrial frogs belies the true diversity of reproductive modes in the Amphibia.

The diversity of reproductive modes is greater among amphibians than in other vertebrates[3]. All orders of the Amphibia include species which deposit eggs in terrestrial environments, and terrestrial development has evolved several times within the order Anura[3].

Deposition of eggs out of water is a major step toward terrestriality among amphibians. Therefore, it is not surprising that most variation in reproductive modes among anurans occurs in tropical environments where there are high levels of atmospheric moisture[3]. A second major step toward terrestriality occurs in those groups which exhibit direct development. The evolution of direct development has been important in the successful invasion of montane environments by amphibians[3]. Direct development occurs in all Australopapuan microhylid frogs[4].

Despite their abundance, little is known about the breeding biology of New Guinean microhylids. Small numbers of large, heavily yolked eggs are concealed in leaf axils, hollow stems, moss clumps or beneath the ground[4,5]. They may be attended by an adult frog, usually a male[4,5]. These frogs generally do not form breeding aggregations (exceptions may be *Asterophrys turpicula*[6] and *Sphenophryne mehelyi*[7]) and may call spasmodically. Thus any observations are fortuitous and valuable.

This note reports observations on the reproductive biology of an unidentified species of *Oreophryne* near Tabubil in the Western Province of Papua New Guinea (5°17′S, 141°12′E). This species was assigned to *Oreophryne* on the basis of its partially webbed feet and the presence of small clavicles[8]. Current confusion surrounding the taxonomy of this genus prevents reliable allocation of our specimens to any particular species.

On the evening of 27.xi.1991 a single specimen was found 3.5 km SSW of Tabubil near a small creek, beneath a closed canopy mid-montane rainforest[9]. At the time of collection it was sitting over a clutch of ten eggs on the underside of a leaf 0.8m from the edge of the creek and 1.5m above the ground (Fig. 1). It was not heard calling, but its sex was determined later by dissection. Only two other species of frog were heard calling in the area (*Rana grisea* and an undescribed member of the *Litoria eucnemis* species-group).

The clutch was arranged in two vertical lines of four and six eggs each. The entire clutch was covered with a thin, transparent, membranous structure which yellowed slightly in alcohol. This layer was quite separate from the jelly-like capsule which surrounded the embryos and, although superficially different, may be homologous to the cord that joins the eggs of congeners for which the eggs are known[8,10,11]. The capsules were 3.8-4.0mm in diameter. The embryos were 3.7-3.8mm long. Each egg contained a very large, ovoid yolk body 3.1-3.2mm in maximum diameter.

Fig. 1. A male *Oreophryne* (SAM R40884) attending a clutch of eggs on the underside of a leaf near Tabubil, Papua New Guinea.

A single egg from this clutch was dissected (Fig. 2). The head (1.1×1.1mm) could be distinguished clearly from the laterally compressed body, which was 2.6mm long. Four limb buds were present, although no differentiation of the digits was apparent. Posterior to the hindlimb buds the tail was strongly compressed laterally, but no vascularisation was apparent in the preserved material. Three cephalic lobes and a pair of pigmented eye spots were clearly visible. A single pair of gill buds were present on each side of the head. Dorsally, two dark lines ran the length of the body, indicating that the neural groove had not yet closed. With the exception of the presence of eye pigmentation and the lack of neural groove closure, these microhylid embryos resemble stage five embryos of the South American leptodactylid, *Eleutherodactylus coqui*[12].

Price[11] illustrated a single, much older embryo of *Oreophryne* from Yapen Island, Irian Jaya. The tail was greatly enlarged and highly vascularised late in development, and presumably plays a role in respiratory gas exchange.

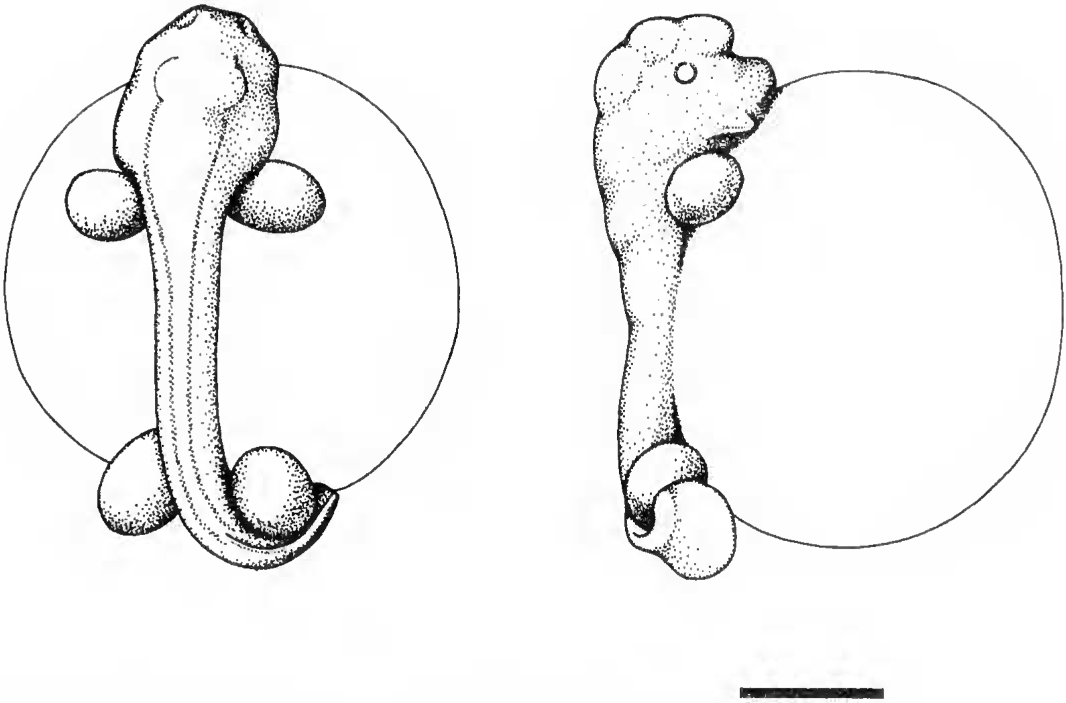

Fig. 2. Dorsal (A) and lateral (B) views of an *Oreophryne* embryo. Scale bar = 1mm.

Measurements of the adult frog were: snout-vent length 22.6mm; head length 7.5mm; head width 7.8mm; eye diameter 3.1mm; eye to naris distance 1.9mm; internarial span 1.8mm; tibia length 10.9mm; foot length 14.2mm. The frog and the clutch it attended have been registered in the South Australian Museum as R40884.

A further two, morphologically identical, male *Oreophryne* were found calling from a cluster of large leaves 3m above the ground, 4m from a sago, swamp amongst mid-montane rainforest 6km SSW of Tabubil at night on 1.xii.1991. Their calls were recorded with a SONY Professional Walkman cassette recorder and an Electret Condenser microphone ECM Z200. Calls were analysed using ULTRASOUND v1.10[13]. The call (Fig. 3) was a biphasic rattle, beginning with a short (0.3-0.4s) series of pulses at 3.2kHz which was followed by a longer (0.7-1.0s) series of pulses at the fundamental and dominant frequency of 2.8kHz, and at 3.2kHz. There were 26 pulses.s^{-1}. Eight calls uttered by two individuals gave a mean call rate of one per 63s. The call length was 1.0-1.4s (mean = 1.1, SD = 0.17).

The breeding behaviour of thirteen of the 83 species of Australopapuan microhylids[14,15,16,17] has been documented[5,10,11,16,18,19,20]. Four to 55 eggs are laid[11]. This reduction in clutch size associated with large, yolked eggs is a common correlate of terrestrial reproduction in amphibians[21].

Males are known to attend eggs in two *Phrynomantis* spp., two *Cophixalus* spp. and three *Sphenophryne* spp.[3,4,18,19] Both males and females attend eggs in two *Oreophryne* spp. and one species of *Cophixalus*[10,20]. Tyler[7] reported aggregations of *Sphenophryne mehelyi* attending their individual clutches of eggs. It is not known how long this association of adult frogs with their clutches may last. The association of frogs with their eggs may function in manipulation of eggs, to provide moisture for terrestrial eggs, in protection against predators or for removal of dead or infected eggs[20].

This report is the first record of any direct developing species laying its eggs on the exposed surface of a leaf. A number of indirect developing frogs from other families are known to deposit eggs on vegetation overhanging water in the neotropics[22,23], Africa[24], Madasgascar[25], the Phillipines[26] and New Guinea[27]. In all of these species the hatching tadpoles fall into the water below. The *Oreophryne* we observed breeding in New Guinea were also found in association with water. This may have been an artefact, however, as we concentrated our searches for frogs around water bodies and did not search the forest surrounding them as thoroughly.

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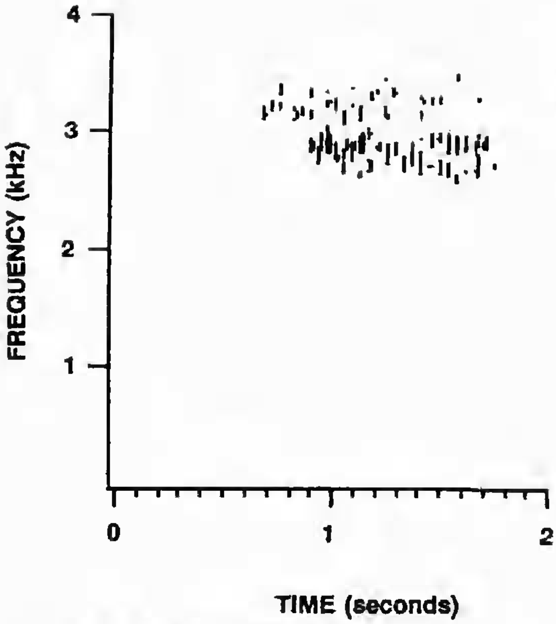

Fig. 3. Sonagram of the call of an *Oreophryne* from 6 km southeast of Tabubil, Papua New Guinea.

assisted with logistic support and transport in New Guinea. Ken Sanderson allowed us to use his software to analyse the call. Nelson Kanem assisted in the field. Margaret Davies and Paula Dempsey made helpful comments on the manuscript. Our work in New Guinea could not have been done without the help of Guy Kula (Department for the Environment, PNG), Rosalyn Busava (Institute of Papua New Guinea Studies) and James Menzies (University of Papua New Guinea).

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